Review article

CED
Clinical and Experimental Dermatology

CPD

Mycetoma: reviewing a neglected disease

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doi:10.1111/ced.13642

Summary

Mycetoma caused by either filamentous fungi (eumycotic) or bacteria (actinomycotic) has recently been recognized by the World Health Organization as a neglected tropical disease. Although mycetoma is preventable and treatable, especially in the early stages, it carries high morbidity and a huge socioeconomic burden. Skin and subcutaneous tissue is affected, with a classic presentation of hard woody swellings, discharging sinuses and presence of grains (containing the causative organism). Variants with swelling without sinuses have also been described. Left untreated it may involve underlying bone and muscle, leading to permanent disability. Common actinomycotic species include *Streptomyces somaliensis*, *Actinomadura madurae*, *Actinomadura pelletieri*, *Nocardia brasiliensis* and *Nocardia asteroides*, while *Madurella mycetomatis*, *Madurella grisea*, *Pseudoallescheria boydii* and *Leptosphaeria senegalensis* are common eumycotic agents. Men are more commonly affected than women, and the leg is the most frequently affected site. Diagnosis in suspected lesions is made with the help of grain examination, microscopy, imaging (radiography, ultrasonography, magnetic resonance imaging) and culture, and more recently by molecular methods such as PCR and molecular sequencing. Molecular sequencing for both fungi and bacteria is important for rapid and correct diagnosis, especially in culture-negative cases. Treatment is long, more successful in actinomycetoma than eumycetoma, and may require a holistic approach comprising antimicrobials, surgery and rehabilitation. Mycetoma can be prevented by simple measures such as wearing protective garments and shoes, especially in rural areas and during outdoor activities.

Introduction

Mycetoma or 'Madura foot' is a chronic granulomatous infection of the skin and subcutaneous tissue, which can be either eumycetoma (fungal) or actinomycetoma (bacterial) (Table 1). Bacterial mycetoma accounts for 60% of cases worldwide, while the rest are fungal.[1] Two mycetomas caused by different agents in the same patient have been reported.[2]

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Conflict of interest: the authors declare that they have no conflicts of interest.

Accepted for publication 11 January 2018

Mycetoma was recently recognized by the World Health Organization as a 'neglected tropical disease'. The primary reason for neglect is that it mainly affects poor populations in remote areas with a lack of trained staff, health facilities, diagnostic tools and treatment. Other factors adding to the neglect are the chronic course of the disease and the poor treatment outcome in eumycetoma.[3]

Although the true incidence of mycetoma is not known, most of the cases fall between latitude 15°S and 30°N, the so-called 'mycetoma belt'. The endemic countries include Sudan, Somalia, Senegal, India, Yemen, Mexico and Venezuela.[2] Although mycetoma is preventable and treatable, especially in the early stages, it carries high morbidity and a huge socioeconomic burden. Left untreated it may involve

Table 1 Causative organisms of mycetoma

Eumycetoma	Actinomycetoma
90% of eumycetomas reported worldwide are caused by only four agents:[16] *Madurella mycetomatis*, (most common), *Madurella grisea*, *Pseudoallescheria boydii*, *Leptosphaeria senegalensis*	The most common species are *Nocardia* spp. (mostly in regions with higher humidity), and include *Nocardia brasiliensis*), *Nocardia asteroides*) and *Nocardia otidiscaviarum*
Other agents *Leptosphaeria tompkinsii*, *Pyrenochaeta romeroi*, *Pyrenochaeta mackinonii*, *Cladophialophora bantiana*, *Cladophialophora mycetomatis*, *Curvularia geniculata*, *Exophiala jeanselmei*, *Phialophora verrucosa*, *Acremonium falciforme*, *Acremonium kiliense*, *Neotestudina rosatii*, *Fusarium moniliforme*, *Fusarium solani*, *Aspergillus nidulans*, *Aspergillus flavus*, *Cylindrocarpon cyanescens*, *Trichophyton* spp., *Microsporum* spp. and *Hormonema* spp.	*Actinomadura* spp., such as *Actinomadura madurae*, *Actinomadura pelletieri*. Also *Streptomyces somaliensis* and *Actinomyces israeli*

underlying bone and muscle, leading to permanent disability.

Causative organisms

Pathogenesis

Mycetoma can be caused by a number of organisms. Common actinomycotic species include *Streptomyces somaliensis*, *Actinomadura madurae*, *Actinomadura pelletieri*, *Nocardia brasiliensis* and *Nocardia asteroides*, while *Madurella mycetomatis*, *Madurella grisea*, *Pseudoallescheria boydii* and *Leptosphaeria senegalensis* are common eumycotic agents.

A variety of pathogen- host- and environment-related factors play a role in the pathogenesis of mycetoma. The initial nonspecific inflammatory response and neutrophil chemotaxsis later become more organized and cellular. T helper (Th)2-like responses [interleukin (IL)-10 and IL-4] have been found in primary lesions and in draining lymph nodes in *S. somaliensis* infection and after stimulation of peripheral blood mononuclear cells by *M. mycetomatis* antigens.[4] Experiments in mice have shown that a protective effect is produced by IgM antibodies, not IgG.[2]

Host genetics play a role; Verwer *et al.* found that the chititriosidase enzyme binds to fungal chitin in the mycetoma grain, accounting for the pathogen-eliminating immune response. A polymorphism resulting in decreased chitotriosidase activity was associated with increased likelihood of eumycetoma.[5] Different pathogens secrete various enzymes, immunomodulators and melanin, which help in their survival in human tissue, evading the first-line immune mechanism. The host reaction to different mycetoma-causing pathogens is similar and produces a similar clinical picture.

Mycetoma cases are seen mainly in areas with hot and dry climates with intermittent short periods of heat and rainfall. The organisms causing mycetoma are found in the soil, and usually enter the human body via a thorn prick or similar trauma.[4]

Clinical presentation

Mycetoma involves the skin and subcutaneous tissue to form the classic triad of a hard woody swelling, painless discharging sinuses and presence of grains, which are colonies of bacteria or fungi and vary in colour and size, depending upon the organism (Fig. 1). The clinical presentation of eumycetoma and actinomycetoma are almost identical, with some subtle differences (Table 2). Other clinical variants include swelling without sinuses, a cystic type and a verrucous plaque type.[2] Farmers and other people living in rural areas are more commonly affected. Although the foot is the most common site of involvement,[6] other reported sites include the arm, forearm, back, head and neck,[7] thorax, scalp, and hand and knee. Males are more frequently affected (sex ratio 3–4 : 1), with age ranging between the third and fourth decades.[1] Left untreated, the infection spreads through the fascial planes and gradually involves the underlying bone and muscle, which makes the infection more resistant to treatment. Pain is usually not present, but local temperature may be raised. The organism may also invade the periosteum and adjacent bones, leading to osteomyelitis. Mycetoma affecting the back may lead to vertebral compression, causing neurological manifestations.[1] Lymphatic spread has been reported in few cases.[8]

Diagnosis and investigations

The presence of the clinical triad described above gives a strong clue towards the diagnosis of mycetoma. Further laboratory and imaging investigations should be

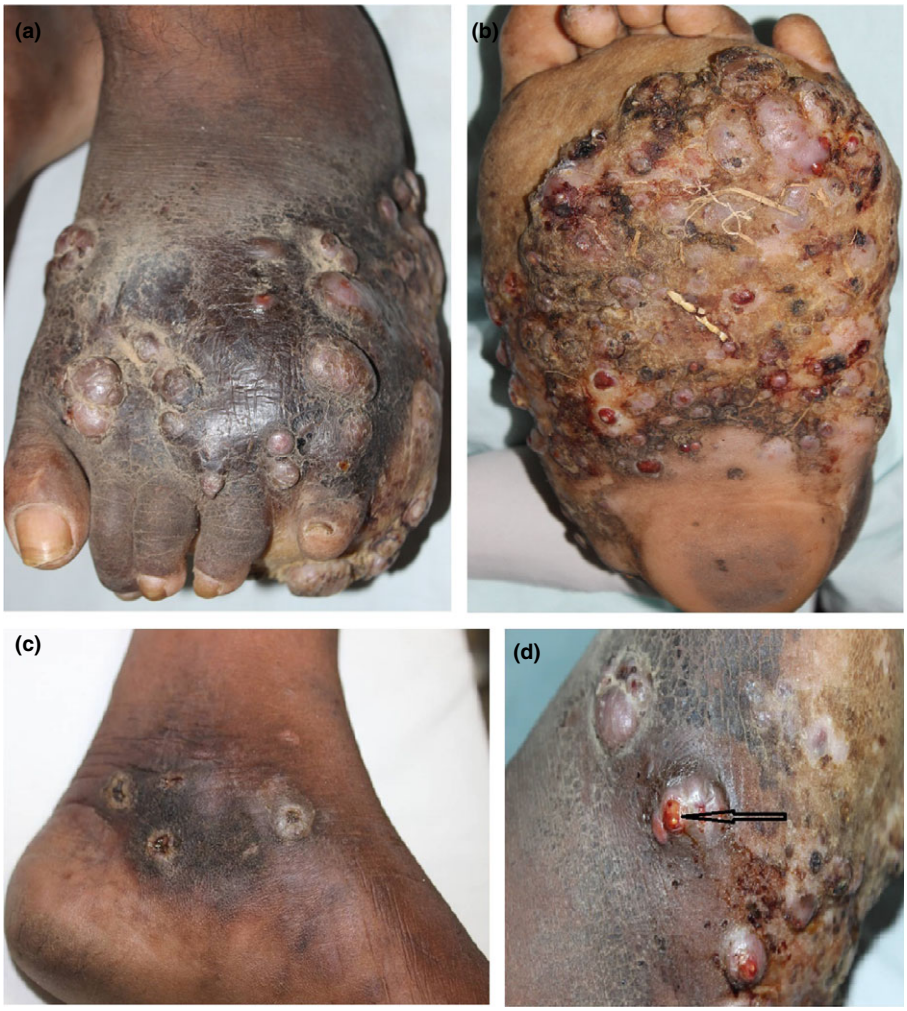

Figure 1 (a–c) Mycetoma of the foot with multiple discharging sinus, (d) closer view of the discharging sinus with yellow grains.

carried out to confirm the presence of causative organisms.

Examination of grains

Grains can be easily visible discharging from the sinuses, or can be collected by use of fine-needle aspiration cytology. Colour, size and consistency of the grains vary for the different causative agents. Large grains are seen with *Madurella* spp., *A. madurae* and *A. pelletieri*, whereas the grains of *N. brasiliensis* and *N. asteroides* are small.[9] Black grains are almost always diagnostic of eumycetoma, while grains of *A. pelletieri* are red. Grains are mostly soft but those of *S. somaliensis* and *M. mycetomatis* can be quite hard.[9] Potassium hydroxide mount of grains will show fungal hyphae, while gram stain and stains for acid-fast bacilli (AFB) are useful in showing filamentous bacteria.

Imaging

Radiographs may be normal, or may demonstrate pathologies such as cortical thinning, hypertrophy, bone cavities and disuse osteoporosis. Eumycotic lesions tend to form a few cavities in bone that are ≥10 mm in diameter, while actinomycotic cavities are often smaller but more numerous.[10]

Ultrasonographic features are helpful in diagnosis and in defining the extent of the lesion (Table 2).

Magnetic resonance imaging (MRI) can also be useful. The MRI finding of The 'dot-in-circle' sign (Fig. 2), seen as tiny hypointense foci within the hyperintense

Table 2 Differences between eumycetoma and actinomycetoma

Characteristic	Eumycetoma	Actinomycetoma
Causative organism	Fungi	Bacteria
Progression	Slowly progressive	Rapidly progressive
Lesions	Well-encapsulated with a clear margin	Diffuse with no clear margin, more inflammatory and destructive
Body part affected	Most commonly the foot,	Most commonly the foot, but also seen on chest, head and abdomen
Sinuses	Few	Many
Colour of grains	Different colours, but mostly white or black	Different colours, but not black
Bone invasion	After a long time	Rapid
Cavities in radiograph	Small in number but large in size, with clear margins	Numerous, small in size with unclear margins
Ultrasonographic features	Thick-walled cavities with hyper-reflective echoes corresponding to grains; more pronounced than in actimomycetoma	Echoes are fine, closely aggregated and commonly settle at the bottom of the cavities
KOH mount of grains	Fungal hyphae seen	No hyphae
Special stains	Periodic-acid–Schiff, Gomori methanamine silver	Gram, AFB. *Nocardia* species are gram-positive and nearly all are weakly acid-fast. while *Actinomadura* grains are Gram-positive and AFB-negative
Medical treatment only	Partial cure or improvement	Improvement in most cases

AFB, acid-fast bacilli; KOH, potassium hydroxide.

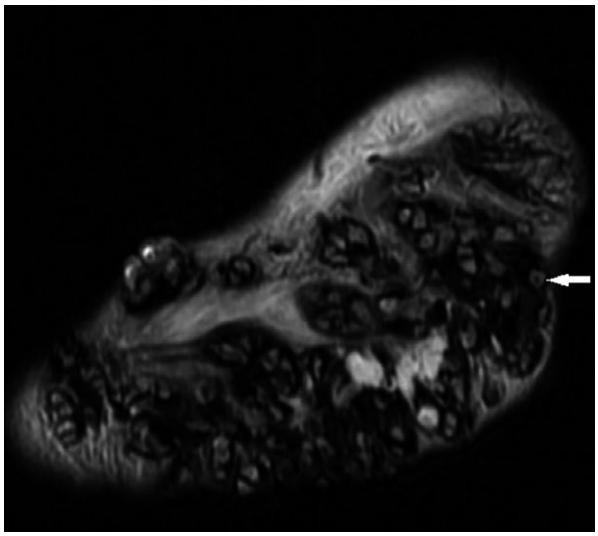

Figure 2 Magnetic resonance imaging scan of the foot showing 'dot in circle' sign.

spherical lesions, was described by Sarris *et al.* as a low-signal matrix representing fibrous tissue, while the granulomata present as high-intensity lesions and the central low-signal focus represents fungal elements (grains).[11]

Histopathology

Histological examination of tissue biopsy reveales suppurative granuloma with mycetoma grains. Periodic-acid–Schiff (PAS), Gomori methenamine silver (GMS) and gram stain along with morphology of grains is helpful in differentiating organisms histopathologically (Fig. 3).

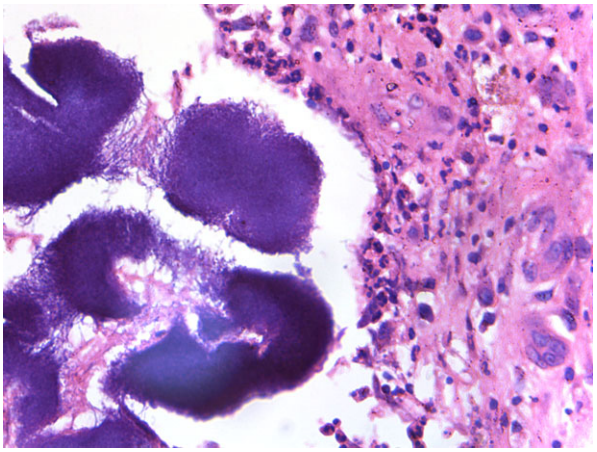

Figure 3 Suppurative granuloma with grains (haematoxylin and eosin, original magnification × 100).

Culture

Using appropriate culture media (Sabouraud or blood agar) colonies of organisms grow after 7–10 days of incubation,[1] although sometimes growth may be

Table 3 Drugs and treatment regimens for actinomycetoma

Regimen	Intensive phase	Maintenance
Welsh regimen[18]	Amikacin 15 mg/kg IM divided into two doses + sulfamethoxazole (35 mg/kg/day) and trimethoprim (7 mg/kg/day) divided into three doses for 21 days. *1–3 cycles at 15-day intervals	Trimethoprim and sulfamethoxazole (7 and 35 mg/kg/day, respectively) continuing for 2 weeks after last cycle
Modified Welsh regimen	Amikacin 15 mg/kg/day in to divided doses + sulfamethoxazole–trimethoprim tablets 35 + 7 mg/kg/day + capsule rifampicin 10 mg/kg/day for 21 days*; 1–3 cycles at 15-day intervals	Sulfamethoxazole–trimethoprim tablets 35 + 7 mg/kg/day + capsule rifampicin 10 mg/kg/day for 3 months
Two step regimen	Crystalline penicillin 1 MU IV every 6 h + gentamicin 80 mg IV every 12 h + cotrimoxazole (trimethoprim–sulfamethoxazole 80/400 mg tablets; two tablets twice daily for 5–7 weeks)	Cotrimoxazole (80/400 mg), two tablets twice daily + amoxicillin tablets, 500 mg thrice daily for 2–5 months after disease becomes inactive
Modified two-step regimen	Gentamicin (80 mg twice daily, IV), and cotrimoxazole (two tablets of 960 mg twice daily) for 4 weeks	Doxycycline (100 mg orally, twice daily), and cotrimoxazole (two tablets of 960 mg twice daily), until 5–6 months after complete healing of all sinuses.
Other drugs used for actinomycetoma Amikacin and dapsone Imipenem Amoxicillin–clavulanate Netilmicin Linezolid DA-7867 (an experimental oxazolidinone): tried *in vivo* against *N. brasiliensis*		
Note: surgical excision may be required along with drugs, especially in localized cases and for lesions with bone and muscle invasion		

IM, intramuscular; IV, intravenous.

delayed for 4–6 weeks. The mycetoma organisms can be identified by their unique morphological and biological activities in culture.

Molecular diagnosis

Molecular diagnosis of the pathogen by direct sequencing of biopsy specimens can be helpful for rapid diagnosis and identification of species for both fungi and bacteria, especially in culture-negative cases. The most commonly used methods are 16s RNA gene sequencing studies for actinomycetes and pan-fungal PCR for eumycetes.[12–14] Other molecular techniques such as loop-mediated isothermal amplification and rolling circle amplification have been successfully used for identification of eumycetoma.[15,16]

Treatment options

There is a wider range of treatment options available for actinomycetoma than eumycetoma, and the latter is more difficult to treat. Surgical treatment is indicated for small, localized lesions and also for large lesions to reduce the organism load.[17]

Various treatment regimens used for actinomycetoma are summarized in Table 3. Treatments for eumycetoma include ketoconazole (400 mg/day), itraconazole (200–400 mg/day), posaconazole (200 mg four times daily), voriconazole 400–600 mg/day, amphotericin B (0.5–1.25 mg/kg per day) and terbinafine (500–1000 mg/day), alone or in any combination.[18] The time taken for remission in actinomycetoma can vary from 3 months to 1 year, whereas eumycetoma requires prolonged treatment ranging from 1 to 3 years.

Conclusion

Mycetoma is seen most commonly in the 'mycetoma belt', and can be caused by various organisms, both bacterial and fungal. The painless nature of the condition leads to delayed presentation, and the lack of diagnostic and treatment facilities in the endemic areas additionally leads to neglect. In general, actinomycetoma responds well to antibiotic treatment, whereas eumycetoma requires a combination of antifungals and surgery and may still have recurrences. Healthcare facilities need to be made available in areas of high burden to detect and treat mycetoma at an early stage.

Acknowledgements

We thank our colleagues at KGMU Lucknow, India: Dr A. Singhai at the Department of Pathology for providing the histopathology images and Dr S. Kumar at the

Department of Radiodiagnosis, for providing the MRI images.

Learning points

- The classic triad of clinical findings for mycetoma consist of hard woody swellings, painless discharging sinuses and presence of grains.
- Actinomycetoma is more inflammatory and progresses more rapidly compared with eumycetoma.
- The MRI finding of a 'dot-in-circle' sign is quite classic for mycetoma.
- Eumycetoma is most commonly caused by *M. mycetomatis*, *M. grisea*, *P. boydii* and *L. senegalensis.*
- Actinomycetoma is mostly caused by *A. madurae*, *S. somaliensis*, *A. pelletieri*, *N. brasiliensis* and *N. asteroides*.
- *Nocardia* species are gram-positive and nearly all are weakly acid-fast, while *Actinomadura* grains are gram-positive and AFB-negative.

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CPD questions

Learning objective

To gain up-do-date knowledge of the causes, features and treatment of mycetoma.

Question 1

Which of the following is the most common organism causing eumycetoma worldwide?

(a) *Madurella mycetomatis.*
(b) *Streptomyces somaliensis.*
(c) *Pseudoallescheria boydii.*
(d) *Leptosphaeria senegalensis.*
(e) *Actinomyces israeli.*

Question 2

Which of the following is true for mycetoma?

(a) Actinomycetoma is less inflammatory than eumycetoma.
(b) Treatment is more successful in eumycetoma than actinomycetoma.
(c) Compared with eumycetoma, actinomycetoma produces more numerous cavities that are small in size with unclear margins in bones.
(d) Black grains are found in Actinomycetoma.
(e) *Nocardia* species are gram-negative.

Question 3

Which one of the following statements about mycetoma grains is true?

(a) Red grains are found in *Madurella mycetomatis*.
(b) The size and colour of grains can give clues towards the aetiological agent in mycetoma.
(c) Presence of grains is necessary to make a clinical diagnosis of mycetoma.
(d) Large grains are seen with *Nocardia brasiliensis*.
(e) Grains present as high-intensity foci on magnetic resonance imaging scans.

Question 4

Which of the following regimens consist of a combination of gentamycin + cotrimoxazole in the intensive phase followed by cotrimoxazole and doxycycline in the maintenance phase for treatment of actinomycetoma?

(a) Welsh regimen.
(b) Modified Welsh regimen.
(c) Two-step regimen.
(d) Modified two-step regimen.
(e) Alternative Welsh regimen.

Question 5

Red grains are produced by which of the following organisms?

(a) *Actinomadura pelletieri*.
(b) *Madurella mycetomatis*.
(c) *Nocardia brasiliensis*.
(d) *Actinomadura madurae*.
(e) *Pseudoallescheria boydii*.

Instructions for answering questions

This learning activity is freely available online at http://www.wileyhealthlearning.com/ced.

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- Reflect on the article
- Register or login online at http://www.wileyhealthlearning.com/ced and answer the CPD questions
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